

Supplemental Material to:

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Myotube-derived exosomal miRNAs downregulate Sirtuin1 in myoblasts during muscle cell differentiation

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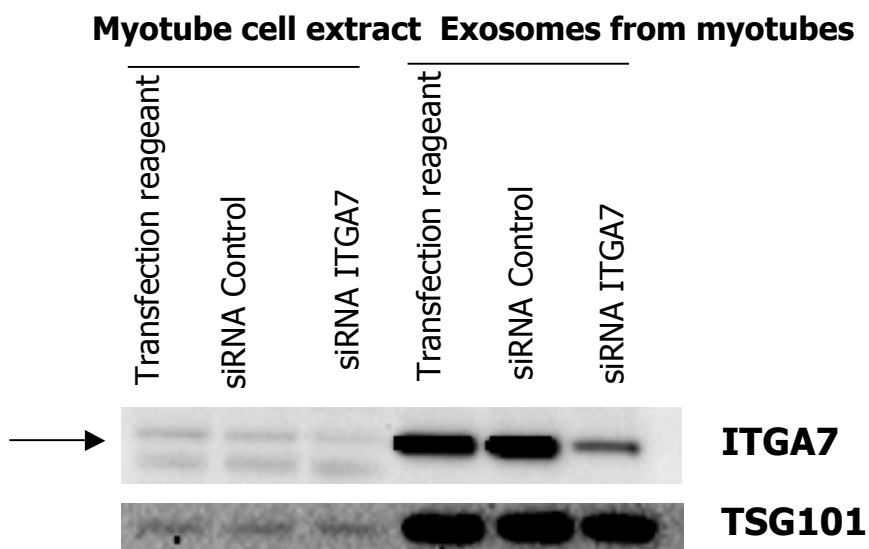
<http://www.landesbioscience.com/journals/cc/article/26808>

Supplementary Table 1: Subsets of microRNAs significantly enriched in exosomes from C2C12 myoblasts and myotubes. MicroRNAs found both in myoblast- and myotube-secreted exosomes are indicated in bold

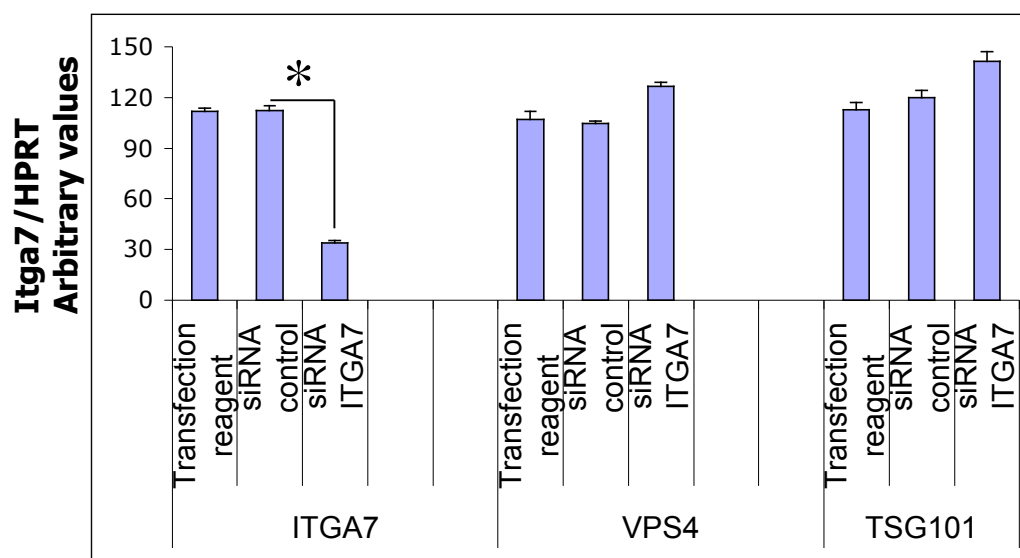
	<i>p</i> -values (Student t- test)	Mean Ct values (myoblasts)	SEM	Mean Ct values (exosomes)	SEM
MYOBLASTS					
mmu-miR-150	0.01111397	29.73193666	0.32636	24.65900316	0.75094
mmu-miR-21*	0.00315996	31.45101495	0.17763	28.63471707	0.30466
mmu-miR-223	0.00553208	33.44534316	0.70878	24.01019832	1.2224
mmu-miR-33*	0.0136745	30.08233222	0.46367	27.52414449	0.34566
mmu-miR-342-3p	0.02606594	25.62269942	0.27824	23.57483118	0.45482
mmu-miR-381	0.0216345	38.26858737	1.51705	31.01236264	0.96933
mmu-miR-383	0.01469653	34.93504206	0.54154	29.48879672	0.97123
mmu-miR-410	0.00508886	37.90206606	0.57847	32.34523645	0.74898
mmu-miR-449c	0.04544337	30.83007554	0.83238	27.55928078	0.31175
mmu-miR-465b-5p	0.00709163	34.18456346	0.37332	29.13328903	0.69173
mmu-miR-467b*	0.01156921	30.84043766	0.33331	27.193937	0.59803
mmu-miR-678	0.02286224	29.32265947	0.16368	25.74720764	0.62098
mmu-miR-704	0.00022615	30.62445365	0.26128	24.2145301	0.35648
mmu-miR-709	0.04501476	17.71281376	0.29611	16.47453443	0.31115
mmu-miR-720	0.02901516	20.71683271	0.4054	18.75519085	0.42565
mmu-miR-760	0.04061276	25.86087273	0.43337	24.18342402	0.33261
mmu-miR-764-5p	0.01131494	30.30920238	0.1246	24.98703855	0.6294
mmu-miR-801	0.04718148	28.11289774	0.13548	20.87447695	1.64649
mmu-miR-804	0.01009687	35.08541503	0.15569	28.90762803	0.70698
mmu-miR-877*	0.00238101	25.28199433	0.29944	22.07136867	0.34774
mmu-miR-99b*	0.01373419	27.48045854	0.31252	25.50626224	0.3473
rno-miR-345-3p	0.0119035	30.80464758	0.3073	28.4669534	0.40575
rno-miR-664	0.00436716	25.94429283	0.14903	24.53249387	0.18185
MYOTUBES					
	<i>p</i> -values (Student t- test)	Mean Ct values (myotubes)	SEM	Mean Ct values (exosomes)	SEM
mmu-miR-150	0.00381949	30.91098081	0.33837	26.82331952	0.10481
mmu-miR-181a-1*	0.02336187	29.00600754	0.37331	26.81617704	0.08372
mmu-miR-183*	0.02628997	26.85614034	0.29741	25.48941596	0.25706
mmu-miR-18a*	0.0087759	30.06166625	0.47057	27.07580574	0.38219
mmu-miR-191*	0.01364324	31.88898231	0.36891	28.68912039	0.57353
mmu-miR-21*	0.00654415	30.09696332	0.39402	27.37946101	0.29787
mmu-miR-342-3p	0.01825107	25.16238333	0.04015	22.53062992	0.37009
mmu-miR-449c	0.01157386	32.22083304	0.38166	27.88304309	0.70514
mmu-miR-467b*	0.00278106	30.16823336	0.3959	26.54188023	0.38422
mmu-miR-542-3p	0.02170116	29.07343157	0.3242	31.37939037	0.48435
mmu-miR-678	0.03636432	28.67922568	0.73793	25.25732209	0.81617
mmu-miR-704	0.0022794	30.44075651	0.67965	24.06070021	0.50225
mmu-miR-706	0.00065083	25.81463724	0.31413	19.51522158	0.13268
mmu-miR-764-5p	0.00173685	28.96795599	0.37713	24.09059206	0.48501
mmu-miR-801	0.0003992	28.5899812	0.33538	21.55200503	0.45802
mmu-miR-804	0.00312153	34.22378442	0.40665	28.9506762	0.60225
mmu-miR-877*	0.01438495	25.16743308	0.58258	21.99930054	0.44677
mmu-miR-878-5p	0.02192809	33.40739242	0.44101	30.84035108	0.5329
rno-miR-345-3p	0.00566844	30.53389364	0.06269	27.44359074	0.27346
rno-miR-463	0.0469897	31.88116981	0.53747	26.81581981	1.32662

Supplementary Figure 1

A



B



Supplementary data 1 : **A_** Endogenous silencing of ITGA7 protein in C2C12 myotubes was validated by western-blot. ITGA7 was concomitantly down-regulated in exosomes as a consequence of its intracellular down-regulation. We controlled the expression of an exosomal marker (TSG101) to validate that siRNA overexpression did not reduce exosome synthesis. **B_** Endogenous silencing of Itga7 in C2C12 myotubes at the mRNA level was also verified by qRT-PCR. mRNA levels of 2 other unrelated genes was also quantified as control (VPS4 and TG101 which are both involved in exosome formation). mRNA level of these control genes was not affected, indicating that alteration of Itga7 mRNA level in siRNA-transfected myotubes is specific.